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*09/86/267*

L4: Entry 1 of 1

File: USPT

May 20, 2003

US-PAT-NO: 6566050

DOCUMENT-IDENTIFIER: US 6566050 B2

TITLE: Enzyme kits and libraries

DATE-ISSUED: May 20, 2003

## INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
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US-CL-CURRENT: 435/4; 435/6

## CLAIMS:

What is claimed:

1. A method of screening clones having DNA recovered from a plurality of species of organisms for a specified enzyme activity, which method comprises: screening for a specified enzyme activity in a library of clones prepared by (i) recovering DNA from a DNA population derived from a plurality of species of organisms; and (ii) transforming a host cell with the DNA of (i) to produce a library of clones which is screened for the specified enzyme activity.
2. The method of claim 1, wherein the DNA is amplified prior to transforming the host cell.
3. The method of claim 1, wherein the DNA is ligated into a vector prior to transforming the host cell.
4. The method of claim 3, wherein the vector comprises at least one DNA sequence capable of regulating production of a detectable enzyme activity from said DNA.
5. The method of claim 3, wherein the vector into which the DNA has been ligated is used to transform a host cell.

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L3: Entry 1 of 1

File: USPT

Mar 4, 2003

US-PAT-NO: 6528249

DOCUMENT-IDENTIFIER: US 6528249 B1

TITLE: Protein activity screening of clones having DNA from uncultivated microorganisms

DATE-ISSUED: March 4, 2003

## INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
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US-CL-CURRENT: 435/4; 435/6

## CLAIMS:

What is claimed is:

1. A method for identifying a protein activity of interest comprising: culturing a gene expression library comprising a pool of expression constructs, each expression construct comprising a vector containing one or more cDNA or genomic DNA fragments, wherein the cDNA or genomic DNA fragments in the pool of expression constructs are derived from a plurality of species of donor eukaryotic organisms, and wherein the cDNA or genomic DNA fragments are each operably-associated with one or more regulatory regions that drives expression of genes encoded by the cDNA or genomic DNA fragments in an appropriate host organism; and detecting the protein activity encoded by the cDNA or genomic DNA fragments.
2. The method of claim 1, wherein the protein activity is enzymatic activity.
3. The method of claim 2, wherein the enzymatic activity is selected from the group consisting of oxidoreductase, transferase, hydrolase, lyase, isomerase, and ligase activity.
4. The method of claim 1, wherein the donor eukaryotic organisms are microorganisms.
5. The method of claim 4, wherein the microorganisms are derived from an environmental sample.
6. The method of claim 4, wherein the microorganisms are a mixed population of uncultured organisms.
7. The method of claim 1, wherein the DNA fragment comprises one or more operons, or portions thereof.
8. The method of claim 7, wherein the operon or portions thereof encodes a complete or partial metabolic pathway.
9. The method of claim 1, wherein the organisms are fungi.

10. The method of claim 1, wherein the organisms are algae.
11. The method of claim 1, wherein the organisms are protozoan.
12. The method of claim 5, wherein the organisms are extremophiles.
13. The method of claim 12, wherein the organisms are thermophiles, hyperthermophiles, psychrophiles, or psychrotrophs.
14. The method of claim 1, wherein the host organism is a bacterial cell.
15. The method of claim 14, wherein the bacterial cell is an E. coli, Bacillus, Streptomyces, or Salmonella typhimurium cell.
16. The method of claim 1, wherein the host organism is a fungal cell.
17. The method of claim 16, wherein the fungal cell is a yeast cell.
18. The method of claim 1, wherein the host cell is a Drosophila S2 or a Spodoptera S9 cell.
19. The method of claim 1, wherein the host organism is an animal cell.
20. The method of claim 19, wherein the animal cell is a CHO, COS or Bowes melanoma cell.
21. The method of claim 1, wherein the host organism is a plant cell.
22. A method for identifying a protein activity of interest comprising: culturing a gene expression library comprising a pool of expression constructs, each expression construct comprising a vector containing one or more cDNA or genomic DNA fragments, wherein the cDNA or genomic DNA fragments in the pool of expression constructs are derived from a plurality of species of donor organisms, and wherein the cDNA or genomic DNA fragments are each operably-associated with one or more regulatory regions that drives expression of genes encoded by the cDNA or genomic DNA fragments in an appropriate host organism, wherein the host organism is a bacterial cell; and detecting the protein activity encoded by the cDNA or genomic DNA fragments.
23. A method for identifying a protein activity of interest comprising: culturing a gene expression library comprising a pool of expression constructs, each expression construct comprising a vector containing one or more cDNA or genomic DNA fragments, wherein the cDNA or genomic DNA fragments in the pool of expression constructs are derived from a plurality of species of donor organisms, and wherein the cDNA or genomic DNA fragments are each operably-associated with one or more regulatory regions that drives expression of genes encoded by the cDNA or genomic DNA fragments in an appropriate host organism, wherein the host organism is a fungal cell; and detecting the protein activity encoded by the cDNA or genomic DNA fragments.
24. A method for identifying a protein activity of interest comprising: culturing a gene expression library comprising a pool of expression constructs, each expression construct comprising a vector containing one or more cDNA or genomic DNA fragments, wherein the cDNA or genomic DNA fragments in the pool of expression constructs are derived from a plurality of species of donor organisms, and wherein the cDNA or genomic DNA fragments are each operably-associated with one or more regulatory regions that drives expression of genes encoded by the cDNA or genomic DNA fragments in an appropriate host organism, wherein the host organism is a plant cell; and detecting the protein activity encoded by the cDNA or genomic DNA fragments.
25. A method for identifying a protein activity of interest comprising: culturing a gene expression library comprising a pool of expression constructs, each expression construct comprising a vector containing one or more cDNA or genomic DNA fragments, wherein the cDNA or genomic DNA fragments in the pool of

expression constructs are derived from a plurality of species of donor organisms, and wherein the cDNA or genomic DNA fragments are each operably-associated with one or more regulatory regions that drives expression of genes encoded by the cDNA or genomic DNA fragments in an appropriate host organism, wherein the host organism is an animal cell; and detecting the protein activity encoded by the cDNA or genomic DNA fragments.